TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A SUBMISSION UNDER 35 U.S.C. 371		ATTORNEY'S DOCKET NO. 1032013-000133
		U.S. APPLICATION NO. (If known) 10/561,844
INTERNATIONAL APPLICATION NO. PCT/FR05/01528	INTERNATIONAL FILING DATE 17 June 2005	PRIORITY DATE CLAIMED 17 June 2004

TITLE OF INVENTION
MONOHYDRATED SODIUM SALT OF S-TENATOPRAZOLE AND THE USE THEREOF IN THERAPY

APPLICANT(S) FOR DO/EO/US
COHEN, Avraham; SCHUTZE, Francois; CHARBIT, Suzy; MARTINET, Frederic; FICHEUX, Herve; HOMERIN, Michel

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. ☐ This is a **FIRST** submission of items concerning a submission under 35 U.S.C. 371.
2. ☒ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a submission under 35 U.S.C. 371.
3. ☐ This is an express request to begin national examination procedures (35 U.S.C. 371(f)). The submission must include items (5), (6), (9) and (21) indicated below.
4. ☐ The US has been elected (Article 31).
5. ☐ A copy of the International Application as filed (35 U.S.C. 371(c)(2))
   a. ☐ is attached hereto (required only if not communicated by the International Bureau).
   b. ☐ has been communicated by the International Bureau.
   c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US).
6. ☐ An English language translation of the International Application as filed (35 U.S.C. 371(c)(2))
   a. ☐ is attached hereto.
   b. ☐ has been previously submitted under 35 U.S.C. 154(d)(4).
7. ☐ Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3))
   a. ☐ are attached hereto (required only if not communicated by the International Bureau).
   b. ☐ have been communicated by the International Bureau.
   c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
   d. ☐ have not been made and will not be made.
8. ☐ An English language translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. ☒ An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. ☐ An English language translation of the annexes of the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

Items 11 to 20 below concern document(s) or information included:

11. ☐ An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
12. ☐ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. ☐ A preliminary amendment.
14. ☐ An Application Data Sheet under 37 CFR 1.76.
15. ☐ A substitute specification.
16. ☐ A power of attorney and/or change of address letter.
17. ☐ A computer-readable form of the sequence listing in accordance with PCT Rule 13*ter*.2 and 37 CFR 1.821-1.825.
18. ☐ A second copy of the published International Application under 35 U.S.C. 154(d)(4).
19. ☐ A second copy of the English language translation of the International Application under 35 U.S.C. 154(d)(4).
20. ☒ Other items or information: A Petition to Make Special and a Declaration in Support of Petition to Make Special

11/5/07 DJ

U.S. APPLICATION NO. (If known)	INTERNATIONAL APPLICATION NO.	ATTORNEY'S DOCKET NO.
10/561,844	PCT/FR05/01528	1032013-000133

The following fees have been submitted:	CALCULATIONS	PTO USE ONLY
21. ☐ Basic National fee (37 CFR 1.492(a)) (1631) $ 300	$ 0	
22. ☐ Examination fee (37 CFR 1.492(c)) If the written opinion prepared by ISA/US or the international preliminary examination report prepared by IPEA/US indicates all claims satisfy provisions of PCT Article 33(1)-(4) (1643) $ 0 All other situations (1633) $ 200	$ 0	
23. ☐ Search fee (37 CFR 1.492(b)) If the written opinion of the ISA/US or the international preliminary examination report prepared by IPEA/US indicates all claims satisfy provisions of PCT Article 33(1)-(4) (1640) $ 0 Search fee (37 CFR 1.445(a)(2)) has been paid on the international application to the USPTO as an International Searching Authority (1641) $ 100 International Search Report prepared by an ISA other than the US and provided to the Office or previously communicated to the US by the IB (1642) $ 400 All other situations (1632) $ 500	$ 0	
TOTAL OF 21, 22 AND 23 =	$ 0	
☐ Additional fee for specifications and drawings filed in paper over 100 sheets (excluding sequence listing in compliance with 37 CFR 1.821(c) or (e) or computer program listing in an electronic medium) (37 CFR 1.492(j)). The fee is $ 250 (1681) for each additional 50 sheets of paper or fraction thereof.		
Total Sheets: 0 -100 = 0 / Extra Sheets: /50 = 0 / Number of each additional 50 or fraction thereof (round up to a whole number): 0 / RATE: x $ 250	$ 0	
☒ Surcharge of $ 130 (1617) for furnishing any of the search fee, examination fee, or the oath or declaration after the date of commencement of the national stage (37 CFR 1.492(h)).	$ 130	
CLAIMS / NUMBER FILED / NUMBER EXTRA / RATE		
Total claims: 0 - 20 = 0 / x $ 50 (1615)	$ 0	
Independent Claims: 0 - 3 = 0 / x $ 200 (1614)	$ 0	
MULTIPLE DEPENDENT CLAIM(S) (if applicable) / + $ 360 (1616)	$ 0	
TOTAL OF ABOVE CALCULATIONS =	$ 0	
☐ Applicant claims small entity status. See 37 CFR 1.27. Fees above are reduced by 1/2	0	
SUBTOTAL =	$ 130	
Processing fee of $ 130 (1618) for furnishing the English translation later than 30 months from the earliest claimed priority date (37 CFR 1.492(i)). +	0	
TOTAL NATIONAL FEE =	$ 130	
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31).$ 40 per property +	0	
TOTAL FEES ENCLOSED =	$ 130	
	Amount to be refunded:	$
	Amount to be charged:	$

a. ☐ A check in the amount of _____ to cover the above fees is enclosed.

b. ☐ Please charge my Deposit Account No. 02-4800 in the amount of _____ to cover the above fees. A duplicate copy of this sheet is enclosed.

c. ☒ The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. 02-4800. A duplicate copy of this sheet is enclosed.

d. ☒ Charge 130.00 to credit card. Form PTO-2038 is attached.

NOTE: Where an appropriate time limit under 37 CFR 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the International Application to pending status.

SEND ALL CORRESPONDENCE TO:

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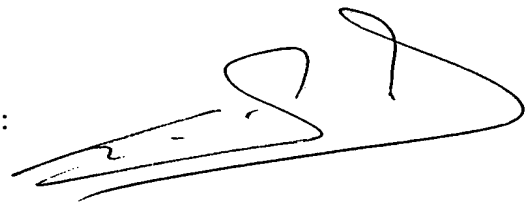
do hereby certify that I am knowledgeable in the French language in which International Patent Application PCT/FR05/001528 was filed, and that, to the best of my knowledge and belief, the English translation is a true and complete translation of the above identified international application as filed.

Signature of Translator : 

Dated this 6th day of December 2005.

MONOHYDRATED SODIUM SALT OF S-TENATOPRAZOLE AND THE USE THEREOF IN THERAPY.

The present invention concerns a tenatoprazole salt, and more particularly a monohydrated salt of the (-) enantiomer of tenatoprazole, or S-tenatoprazole, a method for its preparation as well as its use in human or veterinary therapeutics, namely as proton pump inhibitor (PPI) to treat gastro-oesophageal reflux, digestive bleeding and dyspepsia.

Different sulfoxide derivatives, and notably pyridinyl-methyl-sulfinyl benzimidazoles, have been described in the literature for their therapeutic properties allowing for their use as medicinal products presenting proton pump inhibiting properties to be envisaged, that is to say medicinal products which inhibit the secretion of gastric acid and are useful in the treatment of gastric and duodenal ulcers.

Omeprazole, or 5-methoxy-2-[[(4-methoxy-3,5-dimethyl-2-pyridyl)methyl]sulfinyl]-1H-benzimidazole, described in patent EP 005.129, is one of the first known derivatives of the series of PPIs, possessing gastric acid secretion inhibiting properties, and is widely used as an anti-ulcerative in human therapeutics. Rabeprazole, pantoprazole and lansoprazole can also be found among the other known derivatives of pyridinyl-methyl-sulfinyl benzimidazoles with a similar structure.

Tenatoprazole, or 5-methoxy-2-[[(4-methoxy-3,5-dimethyl-2-pyridyl)methyl]sulfinyl]imidazo[4,5-b]pyridine, is described in Patent No. EP 254.588. It also belongs to the group of drugs classified under the name of "proton pump inhibitors" (PPIs), and can be used for the treatment of conditions such as gastro-oesophageal reflux, digestive bleeding and dyspepsia.

These sulfoxides have an asymmetry at the level of the sulphur atom, and can therefore generally take the form of a mixture (racemic mixture or racemate) of two enantiomers or of

one or the other enantiomer. These enantiomers can classically be used under the form of salts, such as magnesium, potassium or sodium salts, which are generally easier to handle than the bases.

Patent EP 652.872 describes the magnesium salt of esomeprazole, (-) enantiomer of omeprazole, as well as a method for its preparation, the separation of the diastereoisomers and the solvolysis in an alkaline solution. The enantioselective preparation of the (-) enantiomer of omeprazole or of its sodium salts, by oxidation of the corresponding sulphide by a hydroperoxide in the presence of a titanium complex and a chiral ligand is described in patent US 5.948.789. The method described in this patent produces a mixture enriched in one or the other of the (-) and (+) enantiomers, according to the ligand used.

Different formulations have been proposed in order to improve the properties or the activity of PPIs. In the case of omeprazole, for example, PCT application WO 01.28558 describes a stable liquid formulation obtained by forming the sodium or potassium salts *in situ* in solution in polyethylene glycol, by action of a hydroxide on omeprazole. The medicinal product thus formulated can be used in the usual indications of PPIs.

Recent studies have shown that unexpectedly and unlike all the other PPIs (such as, for example, omeprazole or lansoprazole), tenatoprazole possesses a remarkably long duration of action which is the result of a longer half-life in plasma (approximately seven times longer). Indeed, clinical data have shown that tenatoprazole induces a degree of symptom relief and healing of gastric lesions which is superior to those achieved by other PPIs, and which allows for its effective use in the treatment of diseases and conditions such as, for example, atypical and oesophageal symptoms of gastro-oesophageal reflux, digestive bleeding and dyspepsia, as

indicated above. Moreover, it was demonstrated that each of the (+) or (-), or "R" and "S" configuration enantiomers, respectively, contributes differently to the properties of tenatoprazole and the S-tenatoprazole exhibits significantly different pharmacokinetic properties from those of the racemate and of the other enantiomer. S-tenatoprazole is described in French patent application No 2.848.555 published on 18/06/2004.

Studies conducted by the applicant have shown that the monohydrated sodium salt of S-tenatoprazole exhibits unexpected properties which differentiate it from S-tenatoprazole itself, and from other PPIs, and more particularly an excellent solubility which makes it easier to set into its pharmaceutical form and which significantly improves its absorption and the therapeutic efficacy of the medicinal product in which it is contained.

Thus, an object of the present invention is the monohydrated sodium salt of S-tenatoprazole, and the use thereof in human or veterinary therapeutics.

Another object of the present invention is a solution concentrated in monohydrated sodium salt of S-tenatoprazole, and more particularly an aqueous solution at a concentration in monohydrated sodium salt of S-tenatoprazole higher than or equal to 50 g/l, and preferably higher than or equal to 100 g/l.

The present invention also relates to a pharmaceutical composition comprising the monohydrated sodium salt of S-tenatoprazole, substantially free from the (+) enantiomer of R-tenatoprazole, associated to one or more pharmaceutically acceptable excipients and substrates.

A further object of the present invention is the use of the monohydrated sodium salt of S-tenatoprazole in the manufacture of a medicinal product to treat digestive diseases

and conditions where the inhibition of acid secretion must be effective and prolonged to treat, for example, the symptoms and lesions of gastro-oesophageal reflux disease, or digestive bleeding refractory to other PPIs, and especially treat these diseases and conditions in patients receiving multiple drug therapy.

A further object of the present invention is the use of the monohydrated sodium salt of S-tenatoprazole in the manufacture of a drug with a significantly improved rate of healing as well as an increase in the speed of normalization of histological changes of the gastric lesions in animals or humans, which result in a strong decrease in relapses.

The present invention also concerns the use of the monohydrated sodium salt of S-tenatoprazole in the manufacture of a medicinal product with improved pharmacokinetic properties that would allow taking a single dose of medication per day in relevant indications, as indicated hereafter, and particularly in the eradication of *Helicobacter pylori* during the treatment of duodenal ulcer, condition which usually requires two doses (morning and evening) of other PPIs.

Another object of the present invention is an enantioselective method of preparation of the monohydrated sodium salt of S-tenatoprazole, producing the (-) enantiomer salt with a good purity and a satisfactory yield.

The monohydrated sodium salt of S-tenatoprazole can be prepared by enantioselective oxidation of a sulphide of the following general formula (I)

$$A - CH_2 - S - B \qquad (I)$$

where A is a substituted pyridyl nucleus and B an imidazo-pyridyl nucleus, using an oxidising agent in the presence of a vanadium based catalyst and a chiral ligand in a specific sulphide solvent and a specific ligand solvent, according to

the method of preparation described in patent application FR 2.863.611, followed by salification by sodium hydroxide.

In the above-mentioned general formula (I), the pyridyl group A is a 4-methoxy-3,5-dimethyl-2-pyridyl group and B represents a 4-methoxy-imidazo[4,5-b]pyridyl group.

The oxidizing agent used in the method is preferably a peroxide, for example hydrogen peroxide. According to an advantageous method of implementation, highly concentrated hydrogen peroxide is preferably used, higher than 30% for example.

According to the invention, the catalyst may be selected from V oxo-vanadium complex catalysts, and more preferably vanadium acetylacetonate. Such catalysts are commercially available.

A ligand such as a Schiff base derived from a substituted salicylic aldehyde and from a chiral amino-alcohol is preferably used in combination with the catalyst. The choice of the ligand allows for a selective orientation of the reaction towards the desired enantiomer. Thus, the use of 2,4-di-tert-butyl-6-[1-*R*-hydroxymethyl-2-methyl-propylimino)-methyl]-phenol allows for a selective orientation of the oxidation of 5-methoxy-2-[[4-methoxy-3,5-dimethyl-2-pyridyl)methyl]thio]imidazo[4,5-b]pyridine, in order to obtain S-tenatoprazole selectively.

The reaction may be carried out in a solvent, and preferably in a mixture of solvents, in neutral or weakly basic medium, by choosing a specific sulphide solvent and a specific ligand solvent among the group constituted by methanol, tetrahydrofuran, methylene chloride, acetonitrile, acetone and N-methyl-pyrrolidone or toluene, alone or as a mixture. The base possibly used may be a tertiary amine such as pyridine, di-isopropylethylamine or triethylamine. The

oxidation reaction is easily conducted at low or room temperature.

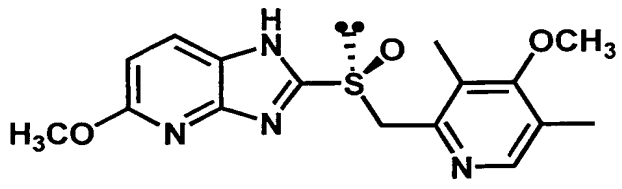
It is particularly advantageous to use the vanadium based catalyst and the ligand in acetonitrile solution, while the sulphide is dissolved in a chlorinated solvent such as dichloromethane, and to combine both solutions before letting the oxidant operate.

More particularly, the oxidation of the sulphide of formula (I) allows for the (-) enantiomer to be obtained, that is, S-tenatoprazole, under good purity and yield conditions by using a vanadium base catalyst associated with a ligand constituted by 2,4-di-tert-butyl-6-[1-*R*-hydroxyméthyl-2-méthyl-propylimino)-methyl]-phenol in acetonitrile solution, while sulphide is dissolved in dichloromethane. Under operating conditions, the ligand and the metallic catalyst form an asymmetric complex where the metal is oxidized by the oxidizing agent.

The oxidation reaction can easily be conducted at low or room temperature, preferably at a temperature comprised between 0 and 10°C so as to facilitate the enantioselectivity.

The sulphide of formula (I) used as starting material is a known product that can be prepared according to several methods described in literature, and for example, according to the methods described in Patents No. EP 254.588 and EP 103.553.

S-tenatoprazole is thus obtained, that is the laevogyre enantiomer of tenatoprazole, and can be represented by the following general formula:

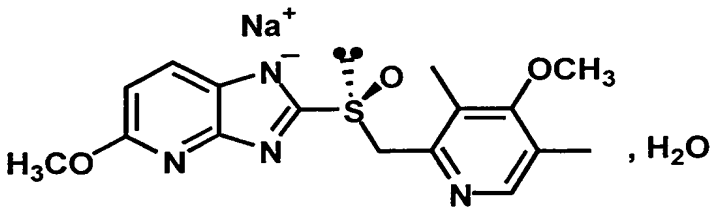


The (-) enantiomer of tenatoprazole, or S-tenatoprazole, corresponds to (-)-5-methoxy-2-[[(4-methoxy-3,5-dimethyl-2-pyridyl)methyl]sulfinyl]imidazo[4,5-b]pyridine, or (-) tenatoprazole. This form can be determined by optical rotation measurements according to standard techniques. Thus, in dimethylformamide and acetonitrile, the angle of optical rotation of (-) tenatoprazole is levorotatory, and its melting point is 130°C (decomposition).

According to an alternative, S-tenatoprazole can also be obtained in a pure optical form from the racemic mixture by well known techniques, using any appropriate method of separation, for example by preparative column chromatography, such as chiral chromatography or high performance liquid chromatography (HPLC). The principle of the chiral chromatography method is based on the difference in affinity existing between (+) and (-) enantiomers and the chiral selector of the stationary phase.

The racemic mixture used as starting material can be obtained using known processes, for example according to the method described in Patent No EP 254.588. Thus, it can be prepared using an oxidizing agent, such as perbenzoic acid, to treat the corresponding sulphide arising from the condensation of a thiol and a pyridine, preferably in the presence of a base such as potassium hydroxide in an appropriate solvent, for example ethanol, under heating. The racemic mixture thus obtained may be separated by HPLC as indicated above.

S-tenatoprazole, obtained according to one or the other above-mentioned methods is then salified in order to obtain a salt with the following formula (II):



In the above formula, the sodium atom may be fixed on the second nitrogen of the imidazopyridyl nucleus close to the sulfoxide group, both isomers being in equilibrium.

The salification is conducted by action of sodium hydroxide on S-tenatoprazole at a temperature comprised between 50 and 70°C, preferably at about 60°C, in a solvent such as water, chloroform, DMSO or a protic solvent, for example methanol or ethanol, then by precipitating the salt obtained after elimination of the solvent. The reaction is preferably conducted under inert atmospheric conditions (nitrogen or argon).

The salt is precipitated according to standard techniques using a solvent miscible with water, where the salt is sparingly soluble, for example a ketone such as acetone and methyl ethyl ketone. The monohydrated salt may be identified by its physico-chemical properties, as indicated further down.

The racemic tenatoprazole salt may be prepared using the same method in order to perform comparative tests, notably solubility tests, with the sodium salt of the isomer.

The thermal analysis measurements and the X-ray diffraction allowed for the characterisation of the structure of the monohydrated sodium salt of S-tenatoprazole, and showed the existence of the monohydrated sodium salt of S-tenatoprazole, which is significantly different from the other forms such as the anhydrous or amorphous forms and the solvates.

Thus, other crystallised phases of the sodium salt may be produced by modifying the crystallisation conditions

(temperature, isolation mode), and the solvents (polarity modulation). For example, the use of dioxane leads to the formation of a perfectly crystallised and characterised solvate of the isomer sodium salt. Nonetheless, the presence of dioxane in the crystalline mesh seems inappropriate for use in pharmaceutics.

The amorphous form, the preparation of which is described in Example 6 here-after, is uncrystallised, instable and difficult to use in pharmaceutical compositions.

Another crystallised phase which can be obtained is the anhydrous sodium salt, described in Example 4 here-after. However, the DVS (Dynamic Vapour Sorption) study revealed the instable nature of this polymorph under usual conditions of relative humidity, leading to the deliquescence of the product. Because of this unstable character, this polymorph is unsuitable for use in pharmaceutics, in particular in usual formulations.

The thermo gravimetric profile of the sodium salt shows that a variable fraction of water (comprised between 1 and 4%) desorbs at low temperature (from 30°C to 50°C) and constitutes a labile and reversible fraction of water. The dehydration of a molecule of water can be observed around 130°C (about 5% of loss of mass). The monohydrated sodium salt was also characterised by DVS (Dynamic Vapour Sorption).

As indicated above, the monohydrated sodium salt of tenatoprazole exhibits excellent solubility properties in water and the major solvents. Thus, the solubility in water ranges from about 140 to 150 g/l at 25°C, and from 240 to 290 g/l at 45°C, which is considerably higher than that of the sodium salt of racemic tenatoprazole (about 18 to 19 g/l), whereas those of racemic tenatoprazole and S-tenatoprazole are lower than 1 g/l.

This result is totally unexpected compared to the solubility of the other well known proton pump inhibitors.

Thus, the monohydrated sodium salt of S-tenatoprazole allows for the preparation of solutions highly concentrated in active medicinal principle, with concentrations higher than 50 g/l, and preferably higher than 100 g/l. For comparison, the racemate sodium salt does not allow for concentrations higher than 19 g/l to be obtained at room temperature.

The monohydrated sodium salt of S-Tenatoprazole exhibits good stability characteristics under normal temperature, pressure and hygrometry conditions. According to environmental and storage conditions, the stoichiometric ratio between the sodium salt and water may evolve and be comprised between 1 and 2. Thus, the water contents corresponding to the sesquihydrated and dihydrated forms may be detected. However, this phenomenon is reversible. The present application relates altogether to the monohydrated sodium salt and to the sesquihydrated and dihydrated sodium salts of S-Tenatoprazole.

A study in the dog showed that the use of the monohydrated sodium salt of S-tenatoprazole allows for a much higher bioavailability to be obtained than with S-tenatoprazole, that is to say a higher concentration (C max) as well as a greater exposure, as measured by the area under the curve of the concentrations according to time (AUC t), for the same dose. Besides, the faster release (Tmax 1.3 hours for monohydrated sodium salt versus 2.5 hours for S-tenatoprazole) allows for therapeutic concentrations to be reached much faster, and thus for the onset of action of the medicinal product to be improved, therefore favouring the possibility of on-demand therapy.

These results are gathered in the following table comparing the monohydrated sodium salt (salt) to basic (free acid) S-tenatoprazole.

dose	T max h	C max $ng.mL^{-1}$	AUC t $ng.h.mL^{-1}$
100 mg/kg (salt)	1.3	183 021	822 785
100 mg/kg (free acid)	2.5	104 751	434 017

The improvement allows for the administered dose to be reduced by a factor of 1.5 to 2, for a comparable exposure. The result is that for a same dose of active principle, the therapeutic efficacy is doubled by the use of the monohydrated sodium salt according to the present invention.

A pharmacokinetic study on dog (n=6) conducted over 4 weeks, comparing the effects of racemic tenatoprazole and of the monohydrated sodium salt of S-tenatoprazole evidenced the original properties of the latter.

The results are gathered in the table below.

dose	T max h	C max $ng.L^{-1}$	AUC t $ng.h.mL^{-1}$
5 mg/kg (salt)	0.5	15 648	42 208
25 mg/kg (salt)	0.5	77 548	148 633
50 mg/kg (salt)	0.7	125 883	323 942
50 mg/kg Racemic tenatoprazole	1.5	50 179	155 592

In this table, the abbreviations have the usual meaning, that is to say that Cmax stands for the maximum plasmatic concentration, Tmax for the time (duration) during which the maximum plasmatic concentration is observed and AUCt for the area under the curve of the plasmatic concentration.

These results were measured on the 28th day of administration.

In this study, the monohydrated sodium salt of S-tenatoprazole, at the doses of 5 mg/kg of weight, 25 mg/kg and 50 mg/kg, and racemic tenatoprazole at the dose of 50 mg/kg, were administered under the form of encapsulated powder.

These results show that the monohydrated sodium salt has a more rapid action (shorter Tmax) than the racemate, whichever the dose used, and provides values of AUC and Cmax that are twice as high for the same dose.

These results were confirmed by a clinical study in man (n=6) during which the patients were successively administered a single dose of: a) capsules of monohydrated sodium salt of S-Tenatoprazole under gastro-resistant form according to a usual technique, b) the same monohydrated sodium salt in powder (non gastro-protected), and c) non salified racemic tenatoprazole, also under the form of capsules containing non gastro-protected powder.

The results obtained are provided in the table below.

Formulation	Cmax $ng.mL^{-1}$	AUCinf $ng.h.mL^{-1}$	T1/2 h
a)	5340	50844	7.81
b)	3199	31223	8.36
c)	2488	21058	7.29

AUC_{inf} is the area under the curve of the plasmatic concentration calculated to the infinite, with extrapolation of the final phase, and $T_{1/2}$ is the plasmatic half-life.

We can therefore observe that the monohydrated sodium salt of tenatoprazole, even non gastro-protected, brings a significant improvement of the parameters.

These results therefore confirm those of the animal studies and demonstrate that the monohydrated sodium salt of

S-tenatoprazole allows for an increase in the exposition (AUC) of about 50% compared to the racemic tenatoprazole. The same goes for the maximum concentration (Cmax).

Thus, the monohydrated sodium salt of S-tenatoprazole not only possesses different pharmacokinetic properties, but also allows for the diminution of the dose by about one third for a similar efficacy.

In the treatment of the conditions listed here-after, the monohydrated sodium salt of S-tenatoprazole can be administered in standard forms adapted to the method of administration chosen, for example *via* the oral or parenteral routes, and preferably *via* the oral or intravenous routes. In particular, the excellent solubility of the monohydrated sodium salt of S-tenatoprazole allows for it to be administered via the intravenous route and thus to ensure the maximal bioavailability of the medicinal product.

The usual formulations of the pharmaceutical technique may be used, for example, it is possible to use tablet or capsule formulations containing the monohydrated sodium salt of S-tenatoprazole as the active principle, or oral solutions or emulsions or solutions for parenteral administration containing the tenatoprazole sodium salt with a standard, pharmaceutically-acceptable substrate.

According to an advantageous form, gastro-resistant granules may also be prepared which can be inserted in a capsule or incorporated in a tablet formulation. The gastro-resistant granules may be prepared by applying a layer of appropriate polymer, such as a cellulosic or methacrylic polymer, for example Eudragit®, on a neutral nucleus carrying a layer containing the active principle.

According to another form particularly adapted to the solubility characteristics of the monohydrated sodium salt of S-tenatoprazole, the nucleus consists in a mixture of a

diluent, for example a cellulosic diluent, a disintegrating agent, and the monohydrated sodium salt of S-tenatoprazole, this nucleus being covered with an enteric film, for example an acetophtalate or methacrylate film.

The disintegrating agent may be a cellulosic polymer, such as cellulose carboxymethyl polymer, for example sodium croscarmellose. The diluent used is preferably an excipient for direct compression, which prevents the use of a wet granulation step. Eudragit® may be used for the enteric coating.

Such a formulation is designed to release the active principle in less than about 5 minutes at pH 6.8, that is to say in the duodenum, after going through the stomach at a more acidic pH.

According to another characteristic, the monohydrated sodium salt has a relative stability in acid medium, which differentiates it from the other proton pump inhibitors. This property allows for the use of the monohydrated sodium salt of S-tenatoprazole in formulations without enteric coating, according to the desired treatment mode. Such formulations present optimised pharmacokinetics and constitute an ideal compromise between the release of the active substance, its immediate action and its relatively small degradation in the stomach. They therefore provide for an additional alternative to the above-described enteric-coated formulations for the practitioner.

The monohydrated sodium salt of tenatoprazole may be used in the manufacture of a medicinal product to treat digestive diseases and conditions where the inhibition of acid secretion must be effective and prolonged to treat, for example, the symptoms and lesions of gastro-oesophageal reflux disease, or digestive bleeding refractory to other PPIs.

The dosage is determined by the practitioner as a function of the patient's state and the severity of the condition. It is generally comprised between 10 and 120 mg, preferably between 10 and 80 mg, more preferably between 15 and 40 mg, of active principle per day.

The excellent solubility of the monohydrated sodium salt of S-tenatoprazole allows for a better absorption of the active principle, and therefore a better bioavailability.

In particular, the bioavailability of the active principle under a form for oral administration, such as tablets or capsules, is close to that obtained by intravenous administration, which leads to the high effectiveness of the product.

The preparation of the monohydrated sodium salt of S-tenatoprazole is described here-after, as well as its original properties, in order to illustrate the present invention, without limiting its scope.

Example 1

Preparation of (S)- (-) tenatoprazole

3 L of methylene chloride and then 360 g of 5-methoxy-2-[[4-methoxy-3,5-dimethyl-2-pyridyl)methyl]thio]imidazo[4,5-b]-pyridine are added in a 5 L flask. The mixture is left under stirring for 30 minutes at room temperature.

700 mL of acetonitrile, 5.22 g of 2,4-di-tert-butyl-6-[1-*R*-hydroxymethyl-2-methyl-propylimino)-methyl]-phenol, and 2.90 g of vanadyl acetylacetonate are dropped one after the other in a 2 L flask. The mixture is kept under stirring at room temperature. After stirring for 30 min, this solution is added to the previous one.

135 mL of hydrogen peroxide at 30% are added to this mixture under stirring for 20 hours at room temperature. After separation of the aqueous phase, the organic phase is washed twice with water, then dried and concentrated under reduced

pressure. 283 g of the desired enantiomer are obtained, with an enantiomeric excess higher than 80% (75% yield). Two successive recrystallisations are performed in a methanol/water or DMF/ethyl acetate mixture and the enantiomer is obtained with an enantiomeric excess higher than 99%.

T_F: 127,5°C

$[\alpha]_D$: -182 ((c 0.1, DMF)

UV spectrum (methanol-water): λ_{max}: 272 nm (ε = 6180), 315 nm (ε = 24877).

Infra-red (KBr) : 3006, 1581, 1436, 1364, 1262 cm^{-1}.

NMR ^{13}C (KOH, reference : sodium 3-(trimethylsilyl)-1-propane-sulfonate) δ (ppm) : 13.2 ; 15.0 ; 56.6 ; 60.8 ; 62.6 ; 107.2 ; 129.5 ; 130.4 ; 131.9 ; 135.1 ; 150.5 ; 151.4 ; 156.9 ; 160.7 ; 163.0 ; 166.6.

MNR ^1H (DMSO d_6, reference: TMS) δ (ppm): 2.20 (s, 6H), 3.70 (s, 3H), 3.91 (s, 3H), 4.69-4.85 (m, 2H), 6.80 (d, *J* 8.5 Hz, 1H), 7.99 (d, *J* 8.5 Hz, 1 H), 8.16 (s, H), 13.92 (s, 1H).

Example 2

Preparation of the monohydrated sodium salt of (S)-(-)-tenatoprazole

1.0 mL of water and 0.6 mL of sodium hydroxide in aqueous solution (5M) are dropped under slow agitation at room temperature in a 50 mL flask equipped with a stirrer, a temperature sensor and a condenser, and containing 1.0 g of S-(-)-tenatoprazole obtained as described in Example 1.

The reaction mixture is heated to 60°C and maintained under stirring for 2.5 hours. An oily liquid is obtained which is cooled down at room temperature before the solvent is eliminated under reduced pressure at 40°C in a rotatory evaporator. After 6 mL of acetone are added under stirring, the pale yellow product precipitates and is collected by

filtration on sintered glass and rinsed in 2.0 mL of acetone or diethyl ether.

After drying at 40°C under reduced pressure for 20 hours, 1.1 g of monohydrated sodium salt of S-tenatoprazole are obtained with a yield higher than 90%.

The monohydrated sodium salt was characterised by thermal analysis and by X-ray diffraction at variable temperature.

Melting point TF: 235°C (capillaries method: Büchi B545 apparatus)

Water content: 5.8% (Karl Fischer)

Enantiomeric excess: higher than 99% (chiral chromatography).

MNR ^1H (DMSO d_6, reference : TMS) δ (ppm): 8.23 (1H, s) ; 7.70 (1H, d, J = 8.4 Hz) ; 6.37 (1H, d, J = 8.4 Hz) ; 4.73 (1H, d, J= 12.9Hz) ; 4.37 (1H , d, J= 12.9Hz) ; 3.82 (3H, s) ; 3.70 (3H, s) ; 2.22 (3H, s) ; 2.21 (3H, s).

Thermogravimetric analysis:

The thermogravimetric analysis is performed using a Netzsch SCA 409 PC/PG thermobalance. The measurements are made in an aluminium crucible between 20°C and 150°C with a heating up speed of 2°/min under nitrogen pressure.

The thermogravimetric profile reveals three successive steps:

- between 10 and 40°C: evaporation, loss of 1.35% of water,
- between 90 and 130°C: dehydration, loss of 4.65% (desorption of a molecule of water),
- between 160 and 230°C: degradation, loss of 9.42%.

Diagram of the X-ray diffraction at variable temperature:

The analysis was conducted with a Siemens D5005 diffractometer (copper anticathode, voltage of 40 kV, intensity of 30 mA, room temperature, measurement range from 3 to 30°, increments between each measurement of 0.04°, measurement time by 4 s).

The measurement data are provided in the table below:

Monohydrated sodium salt of S-tenatoprazole			
Angle 2-Theta °	value of d (Angstrom)	Intensity (Count)	Intensity (%)
5.965	14.80418	508	2.9
6.585	13.41257	17768	100
10.389	8.50818	446	2.5
12.891	6.8615	1352	7.6
13.264	6.66969	670	3.8
15.341	5.77085	676	3.8
17.294	5.12337	507	2.9
19.247	4.60779	444	2.5
19.896	4.45871	1763	9.9
20.925	4.24174	740	4.2
21.6	4.11076	627	3.5
21.824	4.06909	609	3.4
22.316	3.98048	484	2.7
22.885	3.88278	1106	6.2
23.457	3.78939	2731	15.4
25.479	3.49302	637	3.6
26.151	3.40479	864	4.9
26.636	3.34392	709	4
27.506	3.2401	380	2.1
28.32	3.14875	396	2.2
28.526	3.12648	467	2.6
29.708	3.00467	570	3.2

The monohydrated sodium salt was also characterised by DVS (Dynamic Vapour Sorption).

The recordings were performed with a SMS apparatus (Surface Measurement System) with the following characteristics:

- maximum capacity: 1.5 g
- sensitivity: 1.5 µg
- temperature range: 5-48°C
- humidity range: 0-98% RH
- precision: 1% RH

This technique allows for the aptitude of a product to hydrate, dehydrate, solvate and desolvate to be determined by measuring the uptake or the loss of mass according to the controlled atmosphere in terms of water or solvent content at an average temperature.

The results are provided in the following table:

Residual humidity %	Water content (p/p%)	
	Absorption	Desorption
20	3.75	4.40
40	4.45	4.52
60	4.62	4.63
80	4.71	4.71

These results show that the stoichiometry of the monohydrated phase is maintained from 20 to 80% of relative humidity without any deliquescence phenomenon appearing, contrarily to the anhydrous phase (see Example 3). This outlines the excellent stability of the monohydrated sodium salt in presence of humidity.

Under these conditions of strong relative humidity, higher than 80%, the stoichiometry of the sodium salt of S-tenatoprazole may evolve, the number of molecules of water being comprised between 1 and 2. This form, which is also within the scope of the present invention, exhibits an X-ray diffraction at variable temperature diagram similar to the one presented above:

Monohydrated sodium salt of S-tenatoprazole + Second partial hydration			
Angle 2-Theta °	value of d (Angstrom)	Intensity (Count)	Intensity (%)
5.921	14.91531	497	3
6.586	13.40893	16710	100
12.867	6.87461	1252	7.5
13.275	6.6642	675	4
17.269	5.13084	501	3
19.203	4.61808	590	3.5
19.941	4.44894	1967	11.8
20.999	4.22702	946	5.7
23.509	3.78109	1685	10.1
25.511	3.48876	457	2.7
26.262	3.39065	650	3.9
26.727	3.33264	729	4.4
27.544	3.23569	707	4.2
28.602	3.11837	471	2.8
29.765	2.99907	675	4

Example 3

Preparation of the monohydrated sodium salt of S-(-)-tenatoprazole

According to an alternative of the process of Example 2, the monohydrated sodium salt is prepared as follows.

25 mL of chloroform are dropped in a 250 mL three-neck distilling flask equipped with a stirrer, a refrigerant and a temperature sensor. 10 g of S-tenatoprazole obtained as described in Example 1 are added and maintained under stirring until solubilisation in chloroform. The mixture is cooled down on an ice/water bed at 4-5°C, before 150 mL of acetone are added. The mixture is then maintained at 4-5°C.

3.85 g of soda lye (30%) are added under stirring whilst maintaining the temperature at 4-5°C, before the reaction medium is let to return to room temperature (20-25°C) while maintained under stirring for 16 hours. The start of a precipitation can be observed after one hour of contact.

The reaction medium is cooled down to a temperature of 4-5°C on an ice bed and maintained under stirring for 4 hours. After filtration of the reaction medium on sintered glass, the powder is collected and rinsed with 15 mL of previously frozen acetone. After vacuum-drying in an oven at 60°C for one night, about 10 g of product are obtained under the form of monohydrated sodium salt of S-tenatoprazole with a yield higher than 90%.

The characteristics of the salt are identical to those of Example 2.

Comparative Example 4

Preparation of the anhydrous sodium salt of S-(-)-tenatoprazole

Based on the S-(-)-tenatoprazole from Example 1, and using the method described in Example 2, sodium hydroxide in aqueous solution is caused to react on S-tenatoprazole at 60°C

to obtain an oily liquid which is taken up in acetone once the water has been eliminated under reduced pressure, and it has been rinsed and dried. The product obtained is set in suspension in a mixture of methanol / acetonitrile (25/75) at 50°C, then cooled down to 5°C to form a white precipitate which is collected by filtration, working in an environment protected from humidity.

Crystallisation yield: 85%.

The X-ray diffraction at variable temperature diagram performed with a Brüker D5000 type apparatus (copper anticathode, 40 V, 30 mA), provides the following results:

Angle (°) 2-Theta	Value of d	Intensity
6.6	13.3	100
9.5	9.3	1
14.3	6.2	2
15.1	5.9	2
15.9	5.6	2
17.4	5.1	1
18.3	4.8	2
19.9	4.5	8
20.9	4.2	2
21.4	4.1	2
22.1	4.0	1
22.7	3.9	2
22.9	3.9	2
23.9	3.7	2
24.9	3.6	1
26.4	3.4	2
27.2	3.3	2
27.6	3.2	1
29.5	3.0	2
30.5	2.9	1
36.3	2.5	1

DVS characteristics (recorded under the same conditions as in Example 2):

Residual humidity %	Water uptake (p/p%)	
	Absorption	Desorption
0	0.00	3.41
20	0.12	12.09
40	0.25	16.45
60	0.65	19.14
80	24.86	24.86

It can be noted that the anhydrous phase becomes irreversibly deliquescent beyond 60% of relative humidity (residual humidity), contrarily to the monohydrated phase.

Comparative Example 5

Preparation of the 1,4-dioxane solvate / sodium salt of S-(-)-tenatoprazole

Based on the S-(-)-tenatoprazole from Example 1, sodium hydroxide in aqueous solution is caused to react on S-tenatoprazole at 60°C according to the method described in Example 2, in order to obtain an oily liquid which is taken up in acetone once the water has been eliminated under reduced pressure and it has been rinsed and dried.

The product thus obtained is set in suspension in a sufficient volume of 1,4-dioxane at 25°C (1 g for about 100 mL of dioxane). The suspension is concentrated slowly at room temperature for 48 hours and is then filtered to obtain the 1,4-dioxane solvate / sodium salt (1/1) under the form of white powder.

Thermogravimetric analysis:

The thermogravimetric analysis is performed under the conditions described in Example 2.

First, the evaporation is observed.

Second, the desolvation of 1,4-dioxane occurs from 70 to 100°C. The loss of mass in the 3rd and 4th steps justifies the stoichiometry of the 1/1 solvate.

X-Ray diffraction at variable temperature diagram:

The X-ray diffraction at variable temperature is performed with a Brüker D5000 type apparatus (copper anticathode, 40 V, 30 mA) and the results are presented below:

Angle (°) 2-Theta	Value of d	Intensity
7.7	11.5	12
11.5	7.7	39
12.6	7.0	100
13.1	6.8	9
13.3	6.6	8
14.2	6.2	8
14.6	6.1	5
15.2	5.8	17
15.5	5.7	20
17.5	5.1	17
18.2	4.9	15
18.8	4.7	7
20.4	4.4	6
23.3	3.8	57
24.1	3.7	36
25.0	3.6	5
26.5	3.4	7
26.8	3.3	14
34.7	2.6	12
35.3	2.5	13
36.0	2.5	8

Comparative Example 6

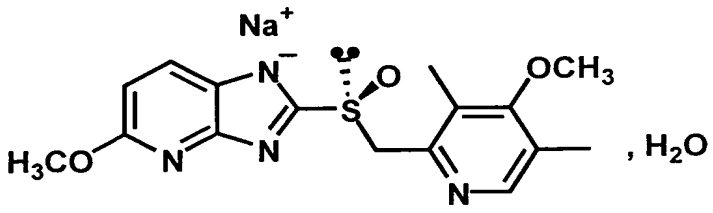
Preparation of the amorphous sodium salt of S-(-)-tenatoprazole

1.0 mL of water and 0.6 mL of sodium hydroxide in aqueous solution (5M) are dropped at room temperature into a 50 mL flask containing 1.0 g of S-(-)-tenatoprazole obtained as indicated in Example 1.

The reaction medium is brought to 60°C and maintained under stirring for 2.5 hours. An oily liquid is obtained which is cooled down at room temperature, before the solvent is eliminated under reduced pressure at 40°C. After 5 mL of water are added under stirring, the amorphous salt precipitates and is collected by filtration. The X-ray diffraction spectrum does not present any diffraction bands.

CLAIMS

1. The monohydrated sodium salt of S-tenatoprazole represented by the general formula (II) here-after:



2. A concentrated solution of monohydrated sodium salt of S-tenatoprazole, wherein the concentration in monohydrated salt is higher than or equal to 50 g/l.

3. A concentrated solution according to claim 2, wherein the concentration in monohydrated salt is higher than or equal to 100 g/l.

4. A pharmaceutical composition comprising the monohydrated sodium salt of S-tenatoprazole according to claim 1, associated to one or more pharmaceutically acceptable excipients and substrates.

5. A composition according to claim 4, wherein it is under the form of unitary doses containing from 10 to 80 mg of active principle.

6. A composition according to claim 5, wherein the unitary dose is comprised between 15 and 40 mg.

7. The use of the monohydrated sodium salt of S-tenatoprazole substantially free from the (+) enantiomer or R-tenatoprazole, for the treatment of digestive diseases.

8. The use of the monohydrated sodium salt of S-tenatoprazole for the manufacture of a medicinal product to treat digestive diseases where the inhibition of acid secretion must be effective and prolonged.

9. The use of the monohydrated sodium salt of S-tenatoprazole for the manufacture of a medicinal product to treat digestive diseases, gastro-oesophageal reflux disease and digestive bleeding in polymedicamented patients.

10. The use of the monohydrated sodium salt of S-tenatoprazole for the manufacture of a medicinal product exhibiting improved pharmacokinetic properties.

11. A method of preparation of the monohydrated sodium salt of S-tenatoprazole, wherein sodium hydroxide is caused to react on S-tenatoprazole at a temperature comprised between 50 and 70°C, and the salt obtained is precipitated after elimination of the solvent.

12. A method according to claim 11, wherein the reaction temperature is of about 60°C.

13. A method according to any of claim 11 and 12, wherein the reaction is conducted in a solvent such as water, chloroform, DMSO or a protic solvent, for example methanol or ethanol.

14. An enantioselective method of preparation of the monohydrated sodium salt of S-tenatoprazole, wherein an enantioselective oxidation is conducted on a sulphide of the following general formulation (I)

$$A - CH_2 - S - B \qquad (I)$$

where A is a 4-methoxy-3,5-dimethyl-2-pyridyl group and B represents a 5-methoxy-imidazo[4,5-b]pyridyl group,
using an oxidising agent in the presence of a vanadium based catalyst and a chiral ligand in a specific sulphide solvent and a specific ligand solvent, followed by salification by sodium hydroxide, in order to obtain the monohydrated sodium salt of S-tenatoprazole.

15. A composition for oral administration of the monohydrated sodium salt of S-tenatoprazole, wherein it consists of a mixture of a diluent, a disintegrating agent and

the monohydrated sodium salt of S-tenatoprazole, this nucleus being covered with an enteric film.

16. A composition according to claim 15, wherein the diluent is a cellulosic diluent.

17. A composition according to claim 16, wherein the diluent is an excipient for direct compression.

18. A composition according to claim 15, wherein the disintegrating agent is a cellulosic polymer, such as a cellulose carboxymethyl polymer.

19. A composition according to claim 18, wherein the disintegrating agent is sodium croscarmellose.

ABSTRACT

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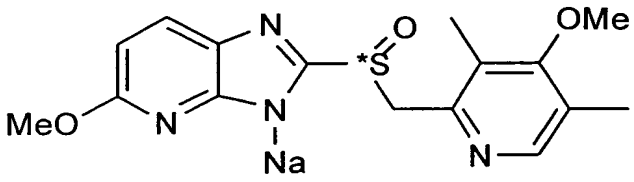
S-tenatoprazole monohydrated sodium salt and the use thereof in therapy.

The present invention relates to S-tenatoprazole monohydrated sodium salt, represented by the following formula :



and the use thereof in therapy for the treatment of digestive diseases.